International Preliminary Examining Authority
European Patent Office
Directorate General 2
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GERMANY

28 April 2006

Sent by fax

Dear Sirs

International Patent Application No. PCT/GB2005/001463
ATHERA BIOTECHNOLOGIES AB
Our Ref: ATHBE/P32968PC

This is a response to the Written Opinion of the International Searching Authority (the "ISA") dated 24 February 2006.

It is to be noted that a Demand for International Preliminary Examination is being filed simultaneously, and that the Demand and this response are both filed within the time limit set down by the communication PCT/ISA/237 from the International Searching Authority, pursuant to Rule 66.1 PCT (i.e. 24 May 2006).

We enclose amended pages 31 and 32, to replace the like numbered pages presently on file.

Unity of Invention – Precautionary Payment of an Additional Examination Fee

In the Written Opinion of the International Searching Authority (ISA), the ISA identified four allegedly non-unitary inventions. Although we disagree with the ISA's allegation of lack of unity, the claims are amended to limit them to the claims categorised as "invention 2" and "invention 4", and we ask that an International Report on Patentability is drawn up in relation to these two inventions, taking into consideration the enclosed amendments and the following arguments.

In particular, it will be clear to the IPEA that, in light of the amendments to the claims, all of the currently pending claims are based on the novel and inventive finding that, *in humans*, antibodies to phosphorylcholine conjugates *provide protection* against cardiovascular diseases such as atherosclerosis, and that *low levels* of these antibodies are *predictive of the risk of cardiovascular disease*. As

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explained in more detail below, the prior art does not teach or even remotely suggest that this would be the case in humans – on the contrary, the prior art teaches that anti-PC antibodies are a risk marker for atherosclerosis. Thus, all claims of the present invention are unitary. Accordingly, only a single examination fee is required.

However, in the present case it is of commercial importance to the applicant to quickly obtain the results of international examination for the claims of both "invention 2" and "invention 4". The applicant has already suffered from considerable delays in the issuance of the International Search Report; it did not issue until nearly *22 months* after the claimed priority date. In view of this already considerable delay, it is most important to the applicant that there is no further delay in obtaining the results of international examination of all claims.

Thus, *merely as a precaution*, we are making the simultaneous payment of an additional examination fee. As the claims are clearly unitary, we request a full refund of this additional precautionary fee.

However, in order to avoid the possibility of yet further delays in the examination of the claims of this application then, in the event that the IPEA, incorrectly, finds that the claims, as amended, lack unity, then we specifically request that the precautionary additional examination fee be used in compliance with Article 34(3)(a) PCT and Rule 68 PCT, to allow the IPEA to continue with the examination of all of the claims of this application without the inevitable delay that would, otherwise, follow from recourse to the need to issue an invitation under Rule 68.2 PCT to pay the additional fee.

In short, for the avoidance of doubt, if the IPEA deems it necessary to invite the applicant to pay an additional fee under Rule 68.2 PCT, then we explicitly waive our entitlement to receive such a communication and this letter is to be taken as an express confirmation that our payment of the additional precautionary fee should be used as payment of the fee required under Rule 68.2 PCT to allow the IPEA to continue with the examination of all of the claims of this application without delay.

In any case, the IPEA will appreciate that the claims of the present application are clearly unitary. Accordingly, we do not expect that any additional payment under Rule 68.2 PCT will be required and, in that case, we request a full refund of the additional precautionary fee.

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If the IPEA has any concerns on this matter, we invite them to contact Andrew Wright at our office, by email (awright@eric-potter.com) or telephone (+44 115 955 22 11) in order to discuss this further.

Amendments

Claim 1 has been amended to exclude the subject-matter that was identified by the ISA as "invention 1". Thus, Claim 1 as amended reads as follows (deletions scored-through) –

> "*1. Use of a pharmaceutical composition comprising ~~at least one phosphorylcholine conjugate, or~~ an antibody preparation, for example a monoclonal antibody, with specificity to a phosphorylcholine conjugate in the manufacture of a medicament for immunisation and treatment of ~~mammals, including~~ humans, against atherosclerosis or an atherosclerotic related disease*".

The same substantive amendments have been made to Claims 2 and 9. Claim 8 has been cancelled, and Claim 9 renumbered as new Claim 8 accordingly.

Claim 10 has been cancelled and is replaced by old claim 16, which has been amended as follows (additions underlined, deletions scored through) –

> "*~~16~~9. Use of a phosphorylcholine conjugate in a method of assessing a human patient's risk of developing or progression of ~~ischemic~~ cardiovascular disease in which the patient's levels of ~~IgM or IgG~~ antibodies reactive with the phosphorylcholine conjugate are assessed, wherein low levels of antibodies reactive with the phosphorylcholine conjugate are predictive of the occurrence of cardiovascular disease in a healthy human patient.*"

The amendments to this claim are based on page 25, lines 10-11 of the present application. Page 6, lines 11-14 provide basis for the deletion of the "IgM or IgG" feature.

New Claims 10 and 11 have been added as follows –

> "*10. The use of Claim 9 wherein the cardiovascular disease is ischemic cardiovascular disease.* "

> "*11. The use of Claim 9 wherein the cardiovascular disease is atherosclerosis.* "

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New Claims 12 and 13 specify that the detected antibody is an IgM or IgG antibody, respectively, as set out in previous Claim 16.

Previous Claims 11-15 have been renumbered as new Claims 14 to 18, and have been converted into the same "use" claim format as new Claim 9.

Background of the Invention

The present invention is in the field of atherosclerosis and the treatment and prevention thereof.

Atherosclerosis is a cardiovascular disease characterised by the formation of atherosclerotic lesions (also known as "plaques") in artery walls.

Prior to the filing of the present application, the etiology of atherosclerosis was known to be multifactorial albeit relatively undefined, but some key mechanistic factors in the development of atherosclerosis were considered to include –

- High levels of low density lipoprotein (LDL) in the plasma of a patient, leading to some LDL molecules becoming trapped in the arterial wall, where they would be modified to form oxidised LDL ("oxLDL"); and

- OxLDL uptake by macrophages, resulting the formation of foam cells. The formation of foam cells was considered to be a key event in atherogenesis, resulting in lesion initiation and progression.

This is explained in more detail in, for example, D4 (paragraph bridging pages 1218-1219).

Overview of our comments on inventive step

The claims of the present invention have been limited to the claims of "invention 2", i.e. a *use of antibodies to conjugated phosphorylcholine* ("PC") to treat or prevent atherosclerosis and its related diseases by administration of the antibodies to a patient. Additionally, the claims have been limited to treatment of humans.

In view of the large number of documents cited by the ISA, our comments below are necessarily rather detailed in nature. However, our position on inventive can be summarised in the following bullet points –

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- The ISA's allegation of lack of inventive step against the claims of "invention 2", based on document D10 as the closest prior art, in combination with documents D11-D13 and D15-D17, fails to identify prior art that alone, or in combination, suggest the treatment or prevention of atherosclerosis in humans by administration of antibodies to a PC conjugate. D10 is totally silent on antibodies to PC-conjugates. Rather, it focuses on endogenous levels of antibodies to oxLDL and is, in fact, suggestive of an athero*genic*, not an athero*protective*, role for these antibodies in humans. There is absolutely no suggestion in D10 that atherosclerosis could be treated or prevented in humans by administration of antibodies to PC-conjugates. None of D11-D13 of D15-D17 address the deficiencies in the teaching of D10.

- The ISA's allegation of failure to solve a problem, based on document D14, is totally without scientific foundation. The ISA appear to have confused the issue of treatment of cancer (D14) with treatment or prevention of atherosclerosis (the present invention). These diseases are not the same and what applies to one disease cannot be shown to apply to the other.

- Other prior art in the filed of atherosclerosis, as identified by the ISA in respect of the other "inventions" defined in the Written Opinion of the ISA, further demonstrate that the claims of the present application are inventive.

- For example, the person skilled in the art is taught by documents D1-D6 that short-term atheroprotective effects can be obtained in mutant mice by the provocation an immune response using *S. pneumoniae* as a vaccine. None of these documents provide any motivation to deviate from these teachings in mice. The cautious and conservative nature of the person skilled in the art means that he would not be motivated to adapt the teaching of D1-D6 to arrive at a method of treating humans by administration of antibodies to PC conjugates.

- A challenge of the immune system with *S. pneumoniae* results in the production of a much broader type and variety of antibodies than would be produced in response to a challenge with PC. Thus the prior art, and the present invention, relate to the production of different antibody populations. This provides a clear indication of an inventive step.

- Moreover, the prior art teaches that the atheroprotective effect caused by vaccination of mutant mice with *S. pneumoniae* is a short-term response, whereas the atherogenesis in humans is a long-term disease that takes decades to develop. The prior art fails to suggest a practical, long-term, method for the treatment of atherosclerosis in any animal, much less in humans.

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- All prior art disclosures are based on observations in a mouse model. The prior art is explicitly concerned that its observations may not be applicable to human systems. In view of these concerns, the person skilled in the art would be extremely sceptical about the possibility of treating atherosclerosis in humans, even in the short-term, in the manner disclosed in the prior art for the treatment of mice, i.e. by vaccination with *S. pneumoniae*, and furthermore the prior art fails to suggest alternative treatments. Prior to the disclosure of the present invention, there would have been no realistic motivation to try to treat or prevent atherosclerosis in humans by manipulation the immune system, and certainly there would have been no reasonable expectation of a successful outcome in humans. This provides a further indication of an inventive step.

- Furthermore, in the present application, the inventors have *surprisingly* found that aPC has a long-term athero*protective* effect in the humans. This *could not have been predicted* from the prior art. This surprising finding further demonstrates that the claims relate to inventive subject-matter.

- Additionally, the prior art relating to atherosclerosis is wholly concerned with the provocation of an immune response in the subject to be treated, i.e. active immunisation. By contrast, the present invention, as defined by the claims as amended, does not require the subject to be vaccinated. On the contrary, the claims relate to a method of passive immunisation. There is nothing in the prior art to motivate the person skilled in the art to attempt to treat or prevent atherosclerosis using a method of passive immunisation. This is an additional indication that the claimed subject-matter possesses an inventive step.

- With respect to the claims identified as "invention 4" by the ISA, i.e. Claims 9-16 as amended, the art cited by the ISA in raising an allegation of lack of inventive step is stated by the ISA to teach that higher levels of anti-PC antibodies have a role in and/or are correlated with the development of atherosclerosis – these teachings are based on the examination of a murine model. By contrast, the claims of "invention 4" as amended are related to a method of determining the risk of a *human* patient developing cardiovascular disease, wherein *low* levels of anti-PC antibodies are predictive of the risk of disease. This is based on the surprising finding by the applicants that, in humans, anti-PC antibodies provide long-term protection against cardiovascular disease, despite the fact that the prior art taught that, in mice, antibodies with the ability to bind to PC were considered to be a risk marker. Accordingly it is abundantly clear that the claims of "invention 4" are inventive.

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In view of the foregoing, it will be readily apparent to the IPEA that that claims of the present application are inventive. Our detailed explanation of these issues is given below.

Detailed Comments on Inventive Step

(a) *Comments in response to the ISA's allegation of lack of inventive step*

In Item V of the Written Opinion of the ISA (see sheets 6-7), the ISA have alleged that the claims of "invention 2" lack an inventive step over D10 in combination with any of D11-D13 or D15-D17. The ISA also suggest that D14 is indicative that the claims of "invention 2" fail to solve a problem.

With respect, we disagree with both of the ISA's allegations, particularly in view of the amendments made to the present claims, which have limited the claimed invention to the *treatment of humans*.

The ISA have identified the claims of "invention 2" as relating to the use of a preparation of an antibody specific for a PC conjugate in the treatment of atherosclerosis or related diseases, and a corresponding method of prophylactic or therapeutic treatment, as defined (in part) by Claims 1 to 3 and 9.

None of documents D10-D17 (or, indeed, any of the documents cited by the ISA) disclose treatment (or prophylaxis) of atherosclerosis by administration of an antibody preparation at all (much less by administration of an antibody preparation with specificity to conjugated PC molecule) and so these claims are correctly considered by the ISA to be novel.

The ISA suggest that document D10 teaches the person skilled in the art that increased anti-oxLDL antibody levels reduce the risk of atherosclerosis in a patient and that the method of treatment (or prophylaxis) of atherosclerosis by administering antibodies to a PC conjugate, as defined by the claims of "invention 2" of the present application, differs from the teaching of D10 only in that the present invention applies these findings (i.e. of high anti-oxLDL antibody levels) to achieve a therapeutic treatment of atherosclerosis, thereby to arrive at an allegation of obviousness.

However, contrary to the suggestions of the ISA, D10 certainly would *not* be understood by the person skilled in the art to be teaching that high levels of anti-oxLDL antibodies would be beneficial for the treatment or prevention of atherosclerosis. On the contrary, although D10 tells the reader that anti-oxLDL antibodies can promote clearance of oxLDL from the blood, it is openly questions

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whether this may actually have a *detrimental* effect on atherogenesis. For example –

- Page 174, 1st column, first paragraph of section entitled "*Discussion*" reports that an increase in anti-oxLDL antibodies cannot be assumed to be atheroprotective because –

  > "*There are studies supporting both pro- and anti-atherogenic roles of immunity to oxLDL*";

- The paragraph bridging pages 175-176 reports that –

  > "*It is an important issue whether oxLDL Ab is atherogenic or anti-atherogenic. Some studies have suggested that it is atherogenic, because lipoprotein immune complex is avidly taken up by macrophages leading to massive intracellular cholesteryl ester accumulation [45, 51]. Consistent with this concept are clinical findings that oxLDL Ab titer is elevated in patients with advanced atherosclerosis [22-24]*".

- Page 176, 1st column, lines 12-14 reports that –

  > "*The enhanced uptake of oxLDL immune complexes as shown in vitro may promote atherogenesis when it occurs in arterial wall*" (emphasis added).

- Page 176, 1st paragraph, lines 27-33 reports that –

  > "*the biological role of immune responses against oxLDL presumably differs in various stages of atherogenesis. Although we showed an inverse relationship between circulating oxLDL and oxLDL Ab levels in the healthy population, the role of oxLDL Ab in atherosclerosis awaits further studies in which the extent of atherosclerosis is evaluated*".

Thus, it is clear that in light of the teaching of D10 the person skilled in the art would consider the effect of anti-oxLDL antibodies on the progression of atherogenesis to be, at best, undetermined, and possibly even atherogenic. Accordingly, the ISA's allegation of lack of inventive step is based on a false premise – it cannot even be established that the skilled person would view high

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levels of anti-oxLDL antibodies as being beneficial to the treatment or prevention of atherosclerosis. Accordingly, D10 fails to motivate the skilled person to seek to increase anti-oxLDL antibody levels in a patient.

We remind the IPEA that the claims have been amended to limit the claims to treatment *of humans*.

D10 appears to draw a distinction between the effect of anti-oxLDL antibodies on atherosclerosis in humans and in animals. See D10, page 172, 1st column, lines 1-14, which reports that –

> *"Case-control studies have shown that serum oxLDL Ab titer was elevated in patients with coronary heart disease [22], early-onset peripheral vascular disease [23] and severe carotid atherosclerosis [24]. Raised titer of oxLDL Ab was predictive of carotid atherosclerosis progression [25]. These results suggest the atherogenic role of oxLDL Ab. However, this concept has been challenged by other recent studies. OxLDL Ab titer did not predict atherosclerotic vascular disease in patients with non-insulin-dependent diabetes [26]. Reduction of oxLDL titer was reported in elderly patients with ischemic stroke [27] and in patients with acute myocardial infarction [28]"* (emphasis added).

This strongly suggests that antibodies to oxLDL are either associated with increased risk of atherogenesis *in humans*, and/or that the antibodies play no role *in humans* in atherogenesis, ischemic stroke or acute myocardial infarction.

By contrast, D10 teaches that *in animals* antibodies to oxLDL may provide an atheroprotective effect. See D10, page 172, 1st column, lines 14-21, which reports that –

> *"...immunisation of hypercholesterolemic rabbits with oxLDL [29] or malondialdehyde-modified LDL [30] was followed by a remarkable suppression of atherosclerosis. Furthermore, immunosuppression with cyclosporin A promoted atherosclerosis in experimental animals [31]. There reports indicate the possible anti-atherogenic roles of immune response to oxLDL"* (emphasis added).

Thus, D10 clearly teaches away from treatment or prevention of atherosclerosis *in humans* by increasing oxLDL levels.

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Even if the above teachings were, incorrectly, taken to be a motivation for the person skilled in the art to attempt to treat or prevent atherosclerosis *in humans* by increasing the levels of antibodies to oxLDL, then it is clear that the skilled person would be motivated to do so by active immunisation using oxLDL or malondialdehyde-modified LDL (see the above-quoted passage in respect of animal tests).

Active immunisation using oxLDL or malondialdehyde-modified LDL is clearly a different method to the claimed mode of *passive* immunisation (i.e. administration of an antibody preparation), and D10 fails to provide any motivation for a method of passive immunisation.

Furthermore, the IPEA will appreciate that active immunisation with oxLDL or malondialdehyde-modified LDL will cause a complex immune response, as these vaccines will present a large number of epitopes to the immune system and, thereby, result in the production of a varied population of antibody responses and other non-antibody based immune responses. By contrast, the claims of the present application relate to the use of a specific class of antibodies, *viz.* antibodies to a PC conjugate. There is nothing in D10 to motivate the person skilled in the art to use this specific class of antibodies to treat or prevent atherosclerosis by administration to a human patient.

It is, therefore, clear that the claims of the present invention are inventive over the disclosures of D10.

Moreover, none of D11 to D13 or D15 to D17 address the deficiencies in the teachings D10 in such a way that would motivate the person skilled in the art to modify the teaching of D10 to arrive at a method of treating or preventing atherosclerosis in a human patient by the administration of antibodies to a PC conjugate.

Document D11 discloses a human Mab Fab, cloned by phage display (see abstract) by screening for an antibody that binds to oxLDL (see page 6, lines 19-21). The identified Mab was named IK17. The ISA states that the antibody is suggested to be useful as a therapeutic agent for targeting atherosclerotic drugs. However, as with D10, there is no disclosure of the specific production of an antibody to a PC conjugate, as defined by the claims of the present application. The molecule oxLDL (i.e. oxidised low density lipoprotein) is a complex, large, molecule that will present a very large number of epitopes to the immune system. Many different types of antibodies could bind to it. This is confirmed by D11, page 2, lines 7-8, which confirms that there are "*various oxidation specific epitopes of OxLDL*". In light of this teaching, the person skilled in the art would

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not be specifically motivated to obtain and use a discrete class of antibodies that bind to PC.

Furthermore, as with D10, there is nothing in document D11 that would lead the person of skill in the art to expect that high levels of antibodies to PC conjugates would be beneficial in humans in the treatment or prevention of atherosclerosis.

Thus, the disclosure in D11 of an Fab that binds to oxLDL, and the possible use thereof to target atherosclerotic drugs, is not motivation to adapt the teaching of D10 in such a way as to arrive at the use an anti-PC antibody in a method of treating or preventing atherosclerosis in a human patient.

The ISA suggest that both of documents D12 and D13 "*disclose the use of a different antigen to elicit anti-atherosclerotic immune response*". Thus, the ISA acknowledge two differences between the method of invention 2 and the disclosure of D12 and D13, namely –

- D12 and D13 teach the reader to use a method *of vaccination* to provoke an immune response in a patient. This is quite different to the administration of a defined antibody preparation to a patient as in the claims of the present application.

- Moreover, D12 and D13 relate to different antigens compared to the present invention. D12 relates to the use of fragments of apolipoprotein B to immunise against ischemic cardiovascular diseases and D13 reports that a vaccine for atherosclerosis can be produced by forming a peptide-aldehyde conjugate, such as a conjugate of apoB100 fragment and MDA (see abstract and Claim 1). These molecules are clearly unrelated to PC conjugates and so it will be apparent to the IPEA that documents D12 and D13 relate to the production of a different population of antibodies compared to the present invention.

In view of these differences, there can be no argument that D12 or D13 would motivate the person skilled in the art to modify the teaching of D10 to arrive at a method of treating or preventing atherosclerosis in a human patient by the administration of antibodies to a PC conjugate. On the contrary, they suggest a completely different method and, therefore, teach away from the claims of the present application.

The ISA reports that D15 discloses the use of a hybridoma for producing an anti-PC antibody, and that the antibody retained its specificity for PC-OVA. The simple disclosure of the production of an anti-PC antibody is not suitable

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motivation for the person skilled in the art to modify the teaching of D10 so as to arrive at a method of treating or preventing atherosclerosis in humans by administration of these antibodies, as claimed by invention 2.

The general teaching of D16 is that antibodies may be modified to comprise an amino acid that has cell adhesive properties (e.g. the fibronectin cell adhesive motif RGDS). According to D16, *any* antibody can be modified in this manner. The authors of D16 happened to choose an anti-PC antibody to exemplify this invention. The modified anti-PC antibody was shown to be capable of mediating cell adhesion (see Example 3 on page 10) and the ability of the antibody to bind to PC was undiminished (as determined by binding to PC-KLH conjugate immobilised on a microtiter plate – see Example 4(2) on page 11). There is nothing in D16 to motivate the skilled person to use anti-PC antibodies in a therapeutic context. Certainly, D16 is silent on the relationship between anti-PC antibodies and atherosclerosis in humans, and so the skilled person, starting from the teaching of D10, would not be motivated to consult D16 and, even if he did so, would take nothing from the teaching of D16.

Document D17 reports on the production of a vaccine against sterols, such as cholesterol, ergosterol and derivatives of ergosterol. This is said to be useful in reducing serum cholesterol levels in order to retard or reduce the severity of atherosclerosis. The ISR refers to the examples of D17 as being of most relevance. Example 1 (page 22) describes several vaccine compositions in which cholesterol is present. In two (compositions (iii) and (v)), PC is also presented but D17 provides no rationale to the inclusion of PC in these compositions. The ISA have not explained why the person skilled in the art would view this unreasoned disclosure of the inclusion of PC as one component of a vaccine composition to be motivation to specifically administer a preparation of antibodies to a PC conjugate to treat or prevent atherosclerosis in humans. On the contrary, the teaching of D17 is to use *active immunisation with sterol-containing* vaccine compositions. This teaches away from the method of administering an anti-PC conjugate antibody preparation as defined by "invention 2" of the present application.

In view of the foregoing, it will be readily apparent to the IPEA that the claims relating to "invention 2" as defined by the ISA (i.e. treatment or prevention of atherosclerosis by administration of antibodies to a PC conjugate), are inventive over D10, particularly since those claims have been limited to the treatment of a human. It will also be apparent that none of D11-D13 or D15-D17 motivate the person skilled in the art to modify the teaching of D10 in such a way as to arrive at the invention as claimed.

Accordingly, the claims of the present invention are clearly inventive over D10

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alone or in combination with any of D11-D13 or D15-D17.

The ISA had also referred to document D14 to substantiate an allegation that the claims of the present invention relate to subject matter that fails to solve a technical problem. The basis of the ISA's allegation is the alleged teaching in D14 that the cellular immune responses of a patient, against cancer can be improved by removal of anti-PC antibodies. On this basis the ISA questions whether administration of anti-PC antibodies may, actually, be harmful to a patient and, thus, fail to solve a problem of providing an efficacious treatment regime.

However, this allegation is totally without foundation. The ISA have no evidence to suggest that what is beneficial or harmful for a cancer patient is, in any way, related to what is beneficial or harmful to a patient with (or at risk of) atherosclerosis. In fact, in D14 it is reported that anti-PC antibodies are suspected to block the ability of macrophages to kill tumour cells (page 6, lines 10-14). The ISA have not provided any evidence to suggest that the killing of tumour cells by macrophages is even remotely linked to the uptake of oxLDL in the progression of atherosclerosis. In short, the ISA's allegation based on document D14 is totally without scientific or evidential foundation and we respectfully request that the allegation be withdrawn.

(b) *It was not obvious to raise an antibody response to conjugated PC*

In the Written Opinion of the ISA, the ISA have alleged that "*Documents D1 to D6 each suggest that vaccines which increase antibodies like EO6 protect against atherosclerosis*" (sheet 6 of the Written Opinion), thereby to draw the conclusion that it is obvious to treat atherosclerosis by using conjugated PC to raise an antibody response.

With respect, we submit that the ISA has extrapolated the teachings of D1 to D6 beyond that which the person skilled in the art would reasonably have done before the priority date of the present invention. The ISA's views are informed by the teaching of the present application and, as such, are based on the impermissible use of hindsight. This is, of course, not an appropriate test for inventive step.

The question that should properly be asked in an assessment of inventive step is, what *would* the skilled person be motivated to try, in view of the prior art? To answer this question, one must first consider the nature of the person skilled in the art. We appreciate that the present application is in the international phase, but we believe that useful guidance on the nature of the person skilled in the art can be taken from the case law of the European Patent Office (EPO). In the field of

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biological sciences (as with the present application) the person skilled in the art is described by the EPO's case law as follows –

> "*The person skilled in the art in the field of biotechnology is well defined by the case law of the boards of appeal. His attitude us considered to be conservative. He would never go against an established prejudice, nor try to enter unpredictable areas nor take incalculable risks*"
>
> (see the EPO's "White Book", i.e. The Case Law of the Boards of Appeal of the EPO, 4th Edition, section I.D.5.1.3, page 111 of the English language version).

In light of the teaching of documents D1 to D6, as relied on by the ISA, it is clear that, to the extent that the skilled person would be motivated to attempt to treat atherosclerosis at all (which we deny as discussed in part (c) below), then he would be motivated to do so by immunisation using the established practice of vaccination with *S. pneumoniae*. Taking each of D1 to D6 in turn, our reasons for this are as follows –

Document D1 reports that, in $LDLR^{-/-}$ mice (an animal model of atherogenesis), the endogenous levels of a particular antibody ("EO6") were high. The author reported that EO6 can block macrophage uptake of oxidised low density lipoprotein (OxLDL) and hypothesised that EO6 might be atheroprotective. To test this theory, the author immunised the atherosclerotic mice with *S. pneumoniae* (since EO6 was found to be identical to an antibody which binds to *S. pneumoniae*), and found that levels of EO6 antibodies increased and atherogenesis was inhibited.

Additionally, the author of D1 immunised mice with a component of OxLDL, termed MDA-LDL. D1 reports that MDA-LDL is not bound by EO6 (i.e. MDA-LDL does not contain a PC-like moiety) but, despite this, still causes increased EO6 antibody production in the atherosclerotic mice, allegedly via the ability of MDA-LDL to cause increased IL-5 levels, which then affects the production levels of various antibodies including EO6.

However, the authors do not show that the atheroprotective effect that is observed in response to immunisation with either *S. pneumoniae* or MDA-LDL can necessarily be specifically attributed to the increased production of the EO6 antibody. EO6 is just one of a very large number of antibody types that show increased production in response to the use of these vaccines. The use of complex vaccines such as *S. pneumoniae* and MDA-LDL will inevitably result in complex

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immune responses, which will even include non-antibody related responses. The person skilled in the art would appreciate that any number of different antibodies, or other immune responses, raised by administration of these vaccines could be responsible for the atheroprotective effect reported in D1 and that successful reproduction of the observed effect might be altered if a different immune response were to be induced.

Although D1 closes by suggesting that "*PC-based immunization could lead to protection against atherosclerosis*" one must interpret this statement in light of the disclosure of the whole contents of the document. D1 does not, in any way, show the person skilled in the art that immunisation with an isolated PC molecule, or an isolated PC conjugate, would lead to an atheroprotective effect. Rather, D1 clearly teaches the skilled person that the desired effect can be achieved by using the complex vaccine of *S. pneumoniae*, or the putative IL-5 modulator MDA-LDL, both of which produce a complex immune response. The skilled person would clearly understand that reference to "PC-based immunisation" was a reference to using *S. pneumoniae* based vaccination, since PC is an immunogenic a component of the *S. pneumoniae* cell wall.

Bearing in mind that the skilled person is "*considered to be conservative*" and would not "*try to enter unpredictable areas nor take incalculable risks*" (see above), it is clear that to the extent that D1 motivates the person skilled in the art to continue to treat atherosclerosis at all (see our comments in part (c) below) then it is motivation to do so using either *S. pneumoniae* or MDA-LDL as vaccines, in order to ensure that the same complex immune responses as reported in D1 are provoked in his subject. The skilled person has no reason to deviate from this teaching – there is certainly no motivation in D1 for him to do so. Indeed, there is a considerable disincentive for him to deviate from the exact teaching of D1, because he would understand that deviation from the identity of the vaccine used could lead to a completely different immune response. The ability of different vaccines to provide an atheroprotective effect would be an "*unpredictable area*" with "*incalculable risks*" of failure. The cautious and conservative skilled person *would*, therefore, continue to use either *S. pneumoniae* or MDA-LDL as vaccines.

Document D2 appears to be the fully published version of D1. Our comments above in respect of D1 also apply to D2. D2 motivate the person skilled in the art to use only either *S. pneumoniae*, or MDA-LDL, to achieve atheroprotective immunisation. It provides no motivation to deviate from this teaching, nor would it have been obvious to the person skilled in the art that one could do so without loosing the reported atheroprotective effect.

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Document D3 does little more than summarise the findings of D2. D3 supports our position that, to the extent that the person skilled in the art would consider D2 to be motivation to try to obtain atheroprotective immunisation, then it is motivation to do so using *S. pneumoniae* as a vaccine, and the person skilled in the art would not consider deviation from this teaching. See D3, page 642, second column, lines 16-19 –

> "*Before seriously considering whether we can prevent atherosclerosis with a pneumococcal vaccine, the function of antibodies to oxLDL must be defined in humans*" (emphasis added).

This clearly shows that, at the time of its publication, the skilled reader of D2 viewed *S. pneumoniae* as the appropriate immunogen to use in attempting to achieve atheroprotective immunisation. There is no motivation to deviate from that teaching, for example in order to use PC alone, much less conjugated PC, nor would it have been obvious to the person skilled in the art that one could do so without loosing the reported atheroprotective effect.

The summary section of D4 suggests that "*new therapeutic options could be developed, such as immunization with oxidation-specific epitopes of oxLDL, or interference with Th1-mediated pathways that lead to section of IFN-γ*". However, there is no specific teaching in D4 of what the identity of the "*oxidation-specific epitopes of oxLDL*" should be. That being the case, the person skilled in the art, having read D4, would consult D1, D2 and/or D3 and determine that, to the extent that it is possible to achieve atheroprotective immunisation, hen one should do so using *S. pneumoniae* as a vaccine. In light of this combination of teachings the person skilled in the art would attempt to use antibodies to a conjugate of PC as a sole therapeutic agent.

Document D5, page 6153, 2nd column, reports that various different anti-PC antibodies are able to bind to atherosclerotic lesions. Page 61554, 2nd column, second paragraph of the section entitled "*Discussion*" reports that two group II anti-PC antibodies were "*derived from secondary immune responses to a T cell-dependent PC-protein conjugate*" (emphasis added) and that although those antibodies did not bind PC alone they "*recognized determinants on OxLDL and atherosclerotic lesions*".

However, the disclosure of the production of antibodies that bind to atherosclerotic lesions, in response to PC-protein conjugate, is not the same as disclosure of an ather*oprotective* response using that PC-protein conjugate. D2, page 741, 2nd column tells the reader that "*T-cell-dependent IgG responses to*

cont/....

phosphorylcholine may lead to other, possibly adverse, effects (such as increased foam-cell formation after uptake of immune complexes by Fc-γ receptors)". D5, Table 1 (page 6152) tells us that the group II antibodies to which it refers are IgG1 and IgG2b type antibodies. Therefore, in view of these two disclosures, the person skilled in the art would not form the conclusion that the disclosure in D5 of an IgG-based "*secondary immune response to a T cell-dependent PC-protein conjugate*" that binds to an atherosclerotic lesion is the disclosure of an athero*protective* method – on the contrary, in view of the teaching in D2 the person skilled in the art would expect the response to *promote* atherosclerosis.

Thus, document D5 is evidence that the person skilled in the art, at the time of filing the present application, would be motivated to *avoid* using conjugated PC, in favour of other more "conventional" vaccines, such as *S. pneumoniae*. In other words, D5 shows that that the prior art *taught away* from the idea of using antibody populations that have been raised in response to PC or PC conjugates in order to provide an atheroprotective effect.

Document D6 is an abstract that does little more than confirm the findings of the earlier papers, i.e. that *S. pneumoniae*, and antigens derived therefrom (in this case, R36A antigen and purified cell wall (C-PS)) can cause an anti-PC antibody response.

D6 suggests that "*vaccination with pneumococcal antigens (or even pneumococcal infections) might lead to immune responses that could affect atherogenesis*". This statement confirms that, to the extent that a person skilled in the art would wish to raise an anti-PC antibody response in order to affect atherosclerosis, then the method of choice would be to vaccinate with *S. pneumoniae*, and antigens derived therefrom. There is no suggestion of using antibodies to PC alone, much less conjugated PC.

Thus, contrary to the comments of the ISA, it will be clear that none of D1 to D6 suggest anything other than *S. pneumoniae* and/or MDA-LDL as a vaccine to treat atherosclerosis.

Bearing in mind that the skilled person is "*considered to be conservative*" and would not "*try to enter unpredictable areas nor take incalculable risks*" (see above), it is clear that D1 to D6 motivate the person skilled in the art to continue to attempt to treat atherosclerosis (albeit only in mice in the short-term as discussed in part (c) below) by using either *S. pneumoniae* (or possibly MDA-LDL) as a vaccine, in order to ensure that the same complex immune responses as reported in D1 are provoked in a subject.

cont/....

The skilled person has no reason to deviate from this teaching – there is certainly no motivation in any of D1 to D6 for him to do so. Indeed, there is a considerable disincentive for him to deviate from the exact teaching of D1, because he would understand that deviation from the identity of the vaccine used could lead to a completely different immune response. Evidence of this is in D5, which teaches that use of a PC-conjugate can lead to the production of antibodies that may be of a type suggested in D2 to *promote* atherogenesis.

The ability of different vaccines to provide an atheroprotective effect would be an "*unpredictable area*" with "*incalculable risks*" of failure. The cautious and conservative skilled person *would*, therefore, continue to use either *S. pneumoniae* or MDA-LDL as vaccines.

The ISA's comments do not explain why the person skilled in the art *would* (rather than merely *could*) deviate from the teachings of D1-D6. Where is the motivation for such deviation? The ISA have, with respect, failed to support their position. There is clearly no motivation in D1 to deviation from the use of *S. pneumoniae* or MDA-LDL as vaccines.

In light of this, it will be readily apparent to the IPEA that it was not at all obvious to use antibodies raised specifically in response to conjugated PC in order to treat atherosclerosis, as claimed by the amended claims of the present application.

On the contrary, the claims of the present application are clearly inventive - they relate to the treatment of atherosclerosis by using of a different antibody population than that suggested by the cited art.

(c) It was not obvious to use S. pneumoniae vaccination (or any other alternative) to treat or prevent atherosclerosis in the long-term in humans

Atherosclerosis is a long-term disease that takes *several decades* of a human's life to develop. See, for example, the opening sentence of D4 –

> "*Atherosclerosis is a chronic disease that begins in fetal life, slowly progresses during childhood and adolescence, and then accelerates in fits and spurts in adult life to result in plaque erosion or rupture, effecting morbid or fatal clinical events*".

Thus, a clinically appropriate method of treatment and prevention of atherosclerosis in humans has to be effective over the course of many years or even decades.

cont/....

The reports in the cited prior art of the atheroprotective effect are solely in respect of *mice* that have mutated genotypes that cause accelerated atherogenesis, by vaccination with *S. pneumoniae* and MDA-LDL. These reports are limited to short-term atheroprotective effect of this process.

Documents D1-D3 are explicit in teaching an atheroprotective effect by immunisation with *S. pneumoniae* in mice. Document D1 does not mention any particular time-scale, but it is clear from Document D2 (which is a full report of D1) that the first study related to a period of only 24 weeks after immunisation (see D2, page 739, 1st column, lines 20-21) and the second to a period of only 16 weeks after immunisation (see D2, page 739, 2nd column, lines 20-21). D3 merely summarises the finding of D2 and so adds nothing further.

However, in apparent contradiction to the above findings, the prior art also reports that increases in the levels of anti-oxLDL antibodies are associated with the progression of atherogenesis.

The ISA have recognised this in their comments on documents D19, D20 and D23 (see sheet 8 of the Written Opinion of the ISA). In particular, the ISA correctly states that "*D23 discloses the role of anti-PC antibodies in atherogenesis*".

D20 clearly shows that anti-PC IgM antibodies significantly increase over the time associated with the development of atherosclerosis in *ApoE*$^{-/-}$ mice (see the table, 4th row from the bottom).

D23 reports that –

> "*These studies document that T15 and EO6 Abs are specifically deposited in lesions of murine atherogenesis*" (page 1736, 2nd column, lines 16-18);
>
> "*Ab titers to oxidation-specific epitopes of oxLDL, including those of the IgM isotype, increase in parallel with the development of atherosclerosis in mice*" (page 1739, 1st column, final paragraph); and
>
> "*our data are most consistent with the hypothesis that during progression of atherosclerosis there is an in vivo expansion of the T15/EO6 clonal set in the ApoE*$^{-/-}$ *mice; preliminary studies support this hypothesis...The finding of marked deposition of T15 idiotype Abs in atherosclerotic lesions (Figure 7) also strongly supports this interpretation*" (page 1739, 2nd column, lines 26-33).

cont/....

So in view of this apparently conflicting information of both atheroprotective and atherogenic roles for anti-oxLDL antibodies in mice, how did the person skilled in the art interpret the role of anti-oxLDL antibodies in the progression of atherogenesis? D19, which is a review article covering D23, provides a strong indication of the views of the person skilled in the art. Specifically, D19 tells the reader that the role of anti-oxLDL antibodies in the development of atherosclerosis may change over time as the atherosclerotic organism ages.

See the following passages of D19 –

> "*ApoE$^{-/-}$ mice, which provide a commonly studied animal model for atherosclerosis, ...[show]...vigorous induction of autoantibodies to oxidised LDL*" (page 1683, 2nd column, 2nd paragraph);
>
> "*In ApoE$^{-/-}$ mice, the development of atherosclerosis is paralleled by increasing titres of antibodies to OxLDL*" (page 1683, final sentence);
>
> "*Although the present authors have previously shown (16, 17) that deliberate immunization of hypercholesterolemic rabbits or mice expressing high levels of OxLDL lessens atherosclerosis, the role of autoantibodies against OxLDL needs more evaluation*" (page 1684, 2nd column, lines 6-13); and
>
> "*Since the anti-PC B-cell repertoire changes during the life of the organism, the participation of natural IgM anti-PC antibodies in the disease process may vary with age*" (page 1684, 3rd column, lines 12-16; emphasis added)

In other words, D19 teaches that the role of anti-oxLDL antibodies in the progression of atherosclerosis in mice may be athero*protective* in the short-term, but athero*genic* in the long-term.

Since atherosclerosis in humans is a long-term, chronic, disease, these prior art documents fail to suggest that a practical method of treatment or prevention of atherosclerosis in humans is possible by raising an antibody response to a *S. pneumoniae* vaccine.

Thus, the person skilled in the art has no expectation of being able to succeed in a long-term treatment of atherogenesis in humans by using *S. pneumoniae* vaccination (or any alternative). This also provides a clear indication of an inventive step.

cont/....

(d) *The prior art is explicitly concerned that the observations of an atheroprotective effect in animal models may not be directly attributable to humans*

All of the prior art disclosures of an investigated relationship between anti-oxLDL antibodies and the development of atherosclerosis cited by the ISA relate to observations made *in mice and/or rabbits*.

The cited prior art makes it absolutely clear that the views of those skilled in the art are that it is *extremely uncertain* as to whether the findings in mice and/or rabbits can be applied to humans.

D2 explicitly suggests that the authors are uncertain as to whether its teachings could be successfully transferred from mice to humans. See page 741, the final paragraph of the "Discussion" section –

> "*Much less is known about the human response ... Although the results outlined in this report suggest a protective effect, this could primarily derive from the fact that the dominant antibody response in mice is IgM. The human immune response is more complex, and available pneumococcal vaccines have not been developed to optimise the IgM response to cell wall polysaccharide*[40]. *Moreover, although the development of IgM responses may be beneficial, T-cell-dependent IgG responses to phosphorylcholine may lead to other, possibly adverse, effects (such as increased foam-cell formation after uptake of IgG-oxLDL immune complexes by Fc-γ receptors)*".

Thus, not only does D2 suggest that the short-term atheroprotective observations from mice may not be applicable to humans, but D2 also further suggests that a similar type of treatment in humans may even be atherogenic. This is a clear disincentive to the person skilled in the art to attempt to treat atherosclerosis in humans by increasing the levels of oxLDL antibodies.

Likewise, D3 also casts doubt on whether the short-term atheroprotective method developed in D2 for mice could be successfully transferred to humans. See D3, page 642, second column, lines 16-19 –

> "*Before seriously considering whether we can prevent atherosclerosis with a pneumococcal vaccine, the function of antibodies to oxLDL must be defined in humans*" (emphasis added).

cont/....

The above-quoted passage of D3 makes it absolutely clear that the person skilled in the art would not "*seriously consider*" attempting to treat or prevent atherosclerosis in humans by the method of D2. This is further evidence that a person skilled in the art, upon reading D2, would have had *no reasonable expectation* of successfully treating atherosclerosis in humans using this technique.

Furthermore, as with D3, document D4 also casts further doubt on whether the short-term atheroprotective method developed in D2, as demonstrated on mice, could be successfully transferred to humans. For example, the "*Summary*" section of D4 (beginning on page 1224 and ending on page 1225) reports that "*the action of the immune system in atherosclerosis is in its infancy and much remains to be learned*", that "*[m]ost of the work done so far has been on experimental animals, mainly mice, and the relevance of these observations to human disease remains to be determined*". The authors state that "*[t]he challenge will be to translate what has been learned already...to human populations*". Again, these passages further support our position that high levels of anti-PC antibodies in humans would not be *expected* by the person skilled in the art to successfully treat or prevent atherosclerosis in humans.

Additionally, as discussed in section (a) above, D10 is quite clear in teaching that there is a distinction between the effect of anti-oxLDL antibodies on atherosclerosis in humans and in animals. D10 strongly suggests that antibodies to oxLDL are either associated with increased risk of atherogenesis in humans, and/or that the antibodies play no role in atherogenesis, ischemic stroke or acute myocardial infarction, whereas in experimental animals D10 teaches that in antibodies to oxLDL may provide an atheroprotective effect. This further supports our position that high levels of anti-PC antibodies in humans would not be *expected* by the person skilled in the art to successfully treat or prevent atherosclerosis in humans.

The inability of a person skilled in the art to follow the teaching of the prior art in such a manner that it is reasonable to expect success (in the sense that one can rationally *predict* a positive outcome, rather than *merely hope* for success) can provide a clear indication of an inventive step. In view of the above documents, the person skilled in the art would have *no reasonable expectation of success* of treating or preventing atherosclerosis in a human by increasing the levels of antibodies to oxLDL, much less antibodies to PC conjugates.

Yet the present application surprisingly demonstrates that human subjects having higher levels of anti-PC antibodies ("aPC"), when tested over a period of ten years, have a lower incidence of cardiovascular disease, whereas low levels of

cont/....

aPC in humans over this same period of time are an indicator of risk of cardiovascular disease (see pages 24-25 of the present application, section entitled "*Study showing protective effect of aPC*").

This is a clear indication that the claims of present application relate to inventive subject-matter.

(e) *None of the atherosclerosis-related prior art suggests a method of administering antibodies*

None of the cited prior art relating to atherosclerosis teaches or even remotely suggests that a suitable mode of treatment is by administration of antibodies.

To the extent that the prior art recognises a connection between immune responses and atherosclerosis, the teaching is to manipulate the patient's immune responses such as by vaccination with *S. pneumoniae* or MDA-LDL. Of course, a patient's immune responses can include non-antibody based immune responses.

As discussed above, the person skilled in the art is described by the EPO's case law as follows –

> "*The person skilled in the art in the field of biotechnology is well defined by the case law of the boards of appeal. His attitude us considered to be conservative. He would never go against an established prejudice, nor try to enter unpredictable areas nor take incalculable risks*"
>
> (see the EPO's "White Book", i.e. The Case Law of the Boards of Appeal of the EPO, 4th Edition, section I.D.5.1.3, page 111 of the English language version).

Moreover, as discussed in section (d) of this letter, the cited prior art was explicitly concerned that the observations made in mice may not be applicable to humans.

In view of the conservative nature of the skilled person, particularly in light of the arts' concerns about the unlikely transference of the murine-based effect to humans, the person skilled in the art would not be inclined to introduce further risks of failure by additional modification of the treatment regime disclosed in the prior art.

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Thus, even if the prior art had motivated the person skilled in the art to attempt to treat or prevent atherosclerosis in humans (which we deny), it is clear that the skilled person would not have further deviated from those prior art teachings to arrive at a method that involves administration of an antibody preparation, much less a preparation of antibodies to PC conjugates. On the contrary, the skilled person would have retained as many features of the disclosed treatment regime as possible.

Thus the prior art teaches away from the invention as defined by the claims of "invention 2". This further supports our position that the claims of the present application possess an inventive step.

(f) The claims of "invention 4" are inventive

Claims 9-18, as amended, relate to the use of a PC-conjugate in a method of assessing a *human* patient's risk of developing or progression of cardiovascular disease by assessing levels of anti-PC antibodies wherein *low levels of anti-PC antibodies are predictive* of the disease.

Thus, two key limitations have been included in Claim 9 –

- The claim is limited to humans; and
- Low levels of anti-PC antibody are stated to be predictive of the risk of cardiovascular disease.

None of the prior art cited by the ISA against invention 4 teach either of the above two key limitations in Claim 9. The ISA have cited D19, D20, D21 and D23. The ISA state that D19 and D20 report that increases in anti-PC antibodies are due to atherosclerosis. Likewise, D23 is stated by the ISA to teach that anti-PC antibodies have a role in atherogenesis. In other words, the ISA's position is that D19, D20 and D23 teach that high levels of anti-PC antibody are disease related. By contrast, the data in the present application show that low levels of anti-PC antibodies are related to an increased risk of cardiovascular disease, i.e. higher levels of anti-PC antibodies *provide protection* against cardiovascular disease. This finding is in *direct contrast* to the teachings of D19, D20 and D23 as specified by the ISA.

The reason for this difference is that the present application relates to data collected *from humans*, whereas D19, D20 and D23 relate to data collected

cont/....

from mice (see section (c) above for a more detailed discussion of the contents of D19, D20 and D23).

It could not have been predicted that the role of anti-PC antibodies in the development of cardiovascular disease would be *entirely opposite* mice and humans. This was a very surprising finding.

D21, as relied upon by the ISA in its allegation of lack of inventive step, does not supplement the deficiencies in the teachings of D19, D20 or D23. On the contrary, according to the ISA's position, it merely teaches that cells expressing an anti-PC antibody can be detected using a PC-albumin conjugate. This would not lead the person skilled in the art to the understanding that, *in direct contrast* to the prior art finding in mice, anti-PC antibodies *provide protection* against cardiovascular disease *in humans*.

Therefore, it is quite clear that the claims of "invention 4" possess and inventive step.

(g) Summary of inventive step

In light of the foregoing comments, it is clear that the claims of the present application posses an inventive step over the prior art.

Any amendment is not to be construed as abandonment of subject matter.

Yours faithfully

SMN

Stephen McNeeney PhD
For and on behalf of Eric Potter Clarkson LLP

ajw/amm

Enc: Amended claims pages 31-32